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Evaluation of the Efficacy of Minocycline Therapy for Staphylococcal Soft-Tissue Infection

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Ten patients with soft-tissue infections due to *Staphylococcus aureus* were treated with minocycline, a semisynthetic tetracycline with potent in vitro antistaphylococcal effects. Serum concentrations averaged three to five times the concentration of minocycline required to inhibit growth of *S. aureus* in vitro. Clearing of the infecting organism was slow (less than 50% of lesions were sterile on day 10 of therapy), but clinical improvement was noted in 8 of 10 patients.

Previous studies from this laboratory have demonstrated that therapy of open soft-tissue lesions infected with *Staphylococcus aureus* provides a convenient clinical model for the investigation of in vivo efficacy of antistaphylococcal antibiotics (7). Disappearance of *S. aureus* from lesions during therapy can be correlated with antibiotic susceptibility, serum antibiotic concentrations, and rapidity of clinical healing. We are reporting results of treatment of staphylococcal soft-tissue lesions in 10 patients with minocycline, a semisynthetic tetracycline with potent antistaphylococcal activity (4, 6, 8, 10, 12).

MATERIALS AND METHODS

Ten patients with soft-tissue lesions infected with *S. aureus* received 11 courses of minocycline. Eight patients had abscesses of the buttocks, sacrum, and lower limb, and two had carbuncles, one on the thigh and one on the forearm. The patients were afebrile and did not have leukocytosis, but did have local discomfort in the affected areas. Minocycline was administered for 8 to 18 days. The average length of therapy was 10 days. In addition, all lesions were treated locally with soaks and debridement when necessary. Cultures were obtained Monday through Friday after initiation of minocycline to evaluate, semiquantitatively, the efficacy of therapy. Cultures were taken with a sterile dry cotton swab applied to the surface of the lesions and subsequently streaked over a mannitol-salt agar plate. The plates were incubated for 48 h at 37 C. The number of colonies was estimated and recorded as: (i) too numerous to count, (ii) 200 or fewer colonies, (iii) 50 or fewer colonies, and (iv) negative. Three colonies were picked from each plate for antibiotic susceptibility testing and phage typing.

Susceptibility to multiple antibiotics was determined by the single disk test of Bauer et al. (1). Minimal inhibitory concentrations (MIC) of both tetracycline and minocycline were determined by the standard twofold broth dilution method in a final volume of 1 ml. A 0.5-ml amount of brain heart infusion broth (Difco) was pipetted into a series of test tubes (1.3 by 10 cm), and 0.5 ml of a solution of the antibiotic was then serially diluted in a twofold manner. The last tube contained no antibiotic and served as a positive control. A 0.4-ml volume of brain heart infusion broth was added to each tube. Lastly, 0.1 ml of a 10^{-2} dilution of an 18-h culture of the *S. aureus* to be tested was inoculated into each tube to make the final concentration of the inoculum 10^{-3} organisms per ml (ca. 10^5 colony-forming units). The MIC was read as that concentration of antibiotic in the last clear tube next to the first tube cloudy with growth after 18 h of incubation at 37 C. Minimal bactericidal concentrations were determined by a further 18 h of incubation of the test at 37 C and read the same way as the MICs. Phage typing was performed by a standard manner (3) with a replicate plate technique previously described (11).

Sera were obtained at various intervals after administration of minocycline. The concentration was determined by the agar well diffusion technique of Bennett et al. with *Bacillus cereus* as the test organism (2).

Minocycline was administered orally and given at an initial dose of 200 mg followed by 100 mg every 12 h. No untoward side effects were noted.

RESULTS

Cultures from the 10 patients grew penicillin-resistant *S. aureus* susceptible to minocycline. Six isolates were resistant to tetracycline. Six were phage group I, two were untypable, one was a group III type 84/85, and one was not typed (Table 1).

All the lesions yielded *S. aureus* too numerous to count when initially cultured. Repeat culture obtained from seven patients after 4 days of therapy revealed that three grew 50 colonies or

TABLE 1. *Susceptibility and phage type of S. aureus*[a]

Minocycline treated	MIC (MBC)[b]		Phage type
	Minocycline	Tetracycline	
1	0.4 (0.4)	125 (250)	80/81
2	0.4 (0.8)	125 (250)	80/81
3	0.4 (0.8)	125 (250)	80/81
4	1.56 (3.12)	31.2 (62.5)	84/85
5	0.8 (0.8)	62.5 (62.5)	52/52A/80/81
6	0.05 (0.2)	0.25 (0.5)	52
7	0.4 (0.8)	125 (250)	80/81
8	0.05 (0.1)	0.25 (0.5)	UNT
9	0.05 (0.1)	0.25 (0.5)	UNT
10	0.06 (ND)	0.12 (ND)	ND

[a] Abbreviations: MIC, minimal inhibitory concentration; MBC, minimal bactericidal concentration; ND, not done; UNT, untypable.

[b] Expressed as micrograms per milliliter.

less but that four still had colonies too numerous to count. By day 10, three patients had negative cultures, one grew less than 50 colonies, and four still yielded *S. aureus* too numerous to count. Patient 9 was not recultured, since he responded rapidly and did not return until after healing was complete. In patient 10 the original untypable *S. aureus* had been replaced with a *S. aureus* 80/81 which was susceptible to minocycline. One other patient was colonized by *S. aureus* type 84/85, whereas the original type 80/81 persisted after 10 days of therapy. Lesions of two patients became colonized by *E. coli* and *Pseudomonas* after the *S. aureus* was cleared, but these gram-negative organisms did not appear to retard healing.

Eight of the ten patients demonstrated progressive slow clearing of the exudate of their lesions and clean granulation tissue. One patient with an unsatisfactory response required treatment with a semisynthetic penicillin. The second patient ultimately healed with soaks after discontinuation of minocycline. One patient with a history of recurrent carbuncles had a rapid response, but relapsed within 4 months and required a second course of minocycline. The *S. aureus* was again susceptible to the antibiotic, treatment was reinstituted, and again resolution was noted.

Serum concentrations were measured in five patients following oral administration of 100 mg of minocycline (Table 2). Blood levels greater than concentrations required to inhibit growth of the infecting organism were recorded up to 9 h; at 11 h the levels were often equal to or lower than the MIC of the infecting organism. There appeared to be no accumulation of the drug as determined by assays obtained later in the treatment period.

DISCUSSION

Previous studies have demonstrated that 80 to 90% of daily cultures obtained from soft-tissue lesions in patients receiving either cloxacillin or oxacillin become negative by day 9 of therapy. In contrast, only 12% of daily cultures obtained from lesions treated locally without systemic antibiotics become negative by day 17 of treatment (7). Minocycline therapy resulted in negative cultures in less than 50% of cultures obtained on day 10. Although it is difficult to compare results of therapy carried out in different time periods, these results are less impressive than those achieved with semisynthetic penicillins.

The enhanced antistaphylococcal activity of minocycline noted in vitro is probably explained by an increased uptake of minocycline by *S. aureus*. In contrast, tetracycline-resistant *S. aureus* accumulate less tetracycline than sensitive strains (8). However, the bactericidal activity of minocycline against *S. aureus* in vitro is slow. Ten to fourteen days are required for concentrations of this agent, maintained at 10 to 50 times the MIC necessary to sterilize a 50-ml flask containing an inoculum of 5×10^6 to 1×10^7 colony-forming units per ml of a susceptible *S. aureus* (5). In contrast to these high concentrations utilized in vitro, serum

TABLE 2. *Serum concentration of minocycline*[a]

Patient	Day of therapy	Concn			
		2 to 3 h	5 to 6 h	7 to 9 h	11 h
1	2		1.8		0.5
	7	2.2		1.2	
2	2		2.6		1.0
	7	2.7		2.0	
	14	4.1		3.8	
3	3	2.6			<0.25[b]
	9		1.8		
4	2	1.3			
5	2	1.45	1.45		<0.25
	6	3.5	3.1		
	13	2.2	1.8		

[a] Concentration was measured at the times indicated after 100-mg oral doses and expressed as micrograms per milliliter.

[b] The lower limit of the assay.

levels of minocycline averaged three to five times the MIC of the isolates. In addition, minocycline is 76% protein bound (9), and the MICs of the infecting organism were determined in serum-free media. The high protein binding and slow bactericidal activity may underlie the slower clearing of *S. aureus* from these open wounds treated with minocycline when compared to the therapy with semisynthetic penicillins. Counteracting these possible disadvantages of minocycline treatment of soft-tissue *S. aureus* infection is the finding that this agent is preferentially concentrated in the skin (9). The clinical improvement, which has also been noted by Cappel and Klastersky in the treatment of surgical wound infections (4) in 8 of 10 patients, may reflect the combination of high skin concentrations of minocycline plus the benefits of local treatment.

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